The South African Journal
of
Medical Laboratory Technology

ORGAN OF THE SOCIETY OF MEDICAL LABORATORY
TECHNOLOGISTS OF SOUTH AFRICA

Vol. 4. No. 3. A QUARTERLY September, 1958

Single Numbers 5/6 Annual Subscription £1/1/-

Printed by Provincial Printing Co. (Pty.), Ltd., 482 Smith Street, Durban, and published by the Society of Medical Laboratory Technologists of South Africa, c/o Central Pathological Laboratory, Private Bag, Jacobs, Natal.

The South African Journal
of
Medical Laboratory Technology

ORGAN OF THE SOCIETY OF MEDICAL LABORATORY
TECHNOLOGISTS OF SOUTH AFRICA

Vol. 4, No. 3 A QUARTERLY SEPTEMBER, 1958

CONTENTS

EDITORIAL

What is a profession?

This question was posed by Dr. Vannevar Bush in a recent talk to the American College of Surgeons, and in an address to the 26th Annual Convention of the American Society of Medical Technologists at Milwaukee in June of this year, Dr. Leonard A. Scheele,* chairman of the Commission on Health Careers, National Health Council, discussed the problem. He quotes Dr. Bush as saying that the members of any true profession are the possessors and custodians of a special field of knowledge acquired by assiduous study. They have an obligation to put this professional knowledge and understanding to use . . . and to do their share in expanding the sum total of what is known.

Now let us consider this last statement. Are we vegetating in South Africa, unwilling to work for our professional status by supporting the professional body in its avowed aims? Do we, each one of us, help to make medical technology a better and more interesting profession to the ultimate benefit of the sick, or are we content to work routine hours on routine jobs and no more? When did you, reading this, address a meeting of your colleagues on recent trends, or, for that matter, even attend a meeting? When did you contribute evne a short letter to this ournal? If you cannot remember, then you are one of the hundreds to whom this editorial is addressed.

Membership of the Society of Medical Technologists of South Africa is not obligatory. We rely on professional solidarity for our support but sometimes we despair for that support. We read through lists of branch officers and find the same names recurring; the same people contribute regularly to this journal. They are mostly the older members. What is happening to the younger ones we do not know. Perhaps they have no pride in their profession.

*Year of the Turning Point, 1958. An address presented before the 26th Annual Convention of A.S.M.T., Milwaukee, Wisconsin, June, 1958, which appears in Vol. 24, No. 4 July-August, 1948) of the American Journal of Medical Technology).

LABORATORY TECHNIQUES IN DIAGNOSTIC VIROLOGY

II. The Isolation of Influenza Virus

by

G. S. TURNER

Smith, Andrewes and Laidlaw (1933) first isolated influenza virus from throat washings by the intranasal inoculation of ferrets and demonstrated neutralising antibodies in the convalescent sera of patients. The same authors in 1934 showed that mice were also susceptible. Smith (1935) first demonstrated that the chick embryo was highly susceptible to influenza virus and laid the foundations of the method which is still extant to-day.

The identification of the virus is based primarily on certain inherent properties which are summarised below:

1. It grows readily in the amniotic sac of the chick embryo (Burnet 1940) and subsequently in the allantoic cavity (Nigg *et al* 1940), yielding high concentrations of virus in the cell free extra embryonic fluids.

2. The virus is readily demonstrated in infected fluids by its capacity to agglutinate the erythrocytes of man, guinea pig or fowl. Hirst (1941) and Clark and Nagler (1943).

3. If virus and specific anti-serum are mixed before the addition of red cells, haemagglutination is inhibited (Hirst, 1942).

Materials and Methods: Perhaps the only special piece of equipment or material not in common use in most bacteriological laboratories is a supply of fertile eggs and some means of drilling their shells. The latter is usually accomplished with a dental drill using a carborundum disc burr. A stirrer motor can be adapted with a flexible shaft to fulfil the same task. The moulded papier mâché packing from egg boxes make excellent egg racks.

Human, guinea pig and fowl erythrocytes may be drawn into Alsevers solution and stored for not more than one week at 0 - 4° C. Triple washed cells suspended in saline to 0.5% (pcv) are prepared as required.

i. *Collection of Specimens:* Garglings are taken from patients as soon after the onset of simptoms as possible.* The sample is kept chilled in ice and transported as quickly as possible to the laboratory.

ii. *Preparation of Specimens:* The garglings are transferred to centrifuge tubes and centrifuged at 3,000 - 5,000 r.p.m. for 30 minutes to deposit food debris, mucus and some of the oral bacterial flora.

The topmost 2 mls. of supernatant fluid is carefully pipetted off and the remainder discarded. To the former is added penicillin and streptomycin to give 100 units and 100 mgm. per ml. respectively. It is important throughout all these procedures that the specimens be kept as cold as possible. A refrigerated centrifuge is ideal, but pre-cooled cups and iced balance water are helpful.

iii. *Inoculation of Eggs:* 9 - 11 day old embryos are generally used in this laboratory although some workers recommend older eggs (Beveridge & Burnett, 1946), the position of the embryo and the airsac are pencil-marked on the shell. Their location is determined by transillumination of the egg in a dark room (an old type microscope lamp is excellent for this purpose), the embryo being identified by its prominent eye-spot.

The shell surface over the air space is swabbed with weak tincture of iodine and a circular cut made with the drill. The cut shell cap is removed with sterile forceps exposing the underlying opaque shell membrane. A drop of sterile liquid paraffin is applied to this membrane in the vciinity of the embryo. The shell membrane immediately becomes transparent, rendering visible the underlying vascular chorio-allantoic membrane and below this the embryo itself in its amniotic sac.

Well flamed forceps, with fine points, are used to sear a hole in the shell membrane and the allantois. Through this opening a portion of amnion is picked up and into the amniotic cavity is injected approximately o.1 ml. of the specimen. Inoculation can be effected with either a 1-inch x 27 gauge needleon a 1. ml. tuberculin syringe or a pasteur pipette drawn to a fine capillary.

The egg shell is finally sealed with cellotape which is sterile on its adhesive surface provided it has not been handled. Five or six eggs are usually inoculated from each specimen since there is often a high traumatic mortality rate.

iv. *Incubation of Eggs:* The eggs are set upright on racks and transferred to the incubator at 37 - 39° C. Incubation is continued for approximately 72 hours. Embryos dying during the first 48 hours of incubation are discarded. An open tray of water should stand in the incubator to keep the humidity high.

v. *Harvesting the Embryonic Fluid:* After 40 - 48 hours incubation 2 or 3 eggs are removed from the incubator and chilled at 0 - 4° in the refrigerator to prevent bleeding when the embryonic fluids are sampled. When amniotic fluid is to be removed, the cellophane cap is lightly flamed and removed with sterile scissors. The shell membrane and chorio-allantois are drawn aside with forceps and enough allantoic fluid removed to expose the amniotic sac (a little of this fluid may be saved). The scanty fluid from the amniotic cavity is removed in a similar manner and stored in a sterile container. Usually no abnormalities are visible either in membranes and fluids or in the external appear-

ance of the embryo itself. If negative results are obtained after 48 hours incubation, the remaining eggs may be harvested after 72 hours.

vi *The Testing of Embryonic Fluids:* 1/5 and 1/50 dilutions of the amniotic fluid are made in normal saline, sufficient of each being prepared to allow 3 x 0.5 ml. volumes to be distributed either into Wasserman tubes or the cups of moulded perspex plates. To one series of dilutions is added 0.5 ml. of 0.5% human group O red cells, to another series 0.5 ml. of 0.5% fowl cells and to a final series 0.5 ml. of 0.5% guinea pig cells. With each series a cell control consisting of 0.5 ml. saline and 0.5 ml. cell suspension must be included. The volumes are arbitrary, scanty yields of fluid may necessitate smaller quantities provided these are not so small that reading becomes difficult.

The mixtures are examined after standing at room temperature for one hour. Positive results are characterised by a granular pattern of cells spread over the curved bottom of the tube or cup. Negative results are shown by a compact circumscribed button of cells.

The inclusion of saline controls is necessary since auto-agglutination occasionally occurs with some batches of cells. The cells from three species are used because some influenza types undergo a phasic variation characterised by an affinity for one particular red cell species. Burnet and Bull (1943) have described this transition from the original (O) to the derived (D) phase as O—>D variation. Thus in the O phase it more readily agglutinates the cells of its original (human) host than those of guinea pig and fowl. On adaptation to its new host (chick) this order is reversed. In primary isolation, therefore, it is obviously necessary to include these three red cells species to avoid missing variants. Non-specific agglutination occurs with some samples of amniotic fluid, particularly concentrated specimens; this rarely exceeds a dilution of 1/10, however, and is the reason for including two dilutions in the test.

vii. *Passage:* Completely negative results in first passage material do not exclude the presence of virus. Thus amniotic fluid showing no haemagglutination may be passaged twice more in a further group of embryos in the manner previously described. It is rarely of use to exceed three passages with negative material. Positive samples are diluted at least one hundred fold in nutrient broth and inoculated into the allantoic cavity of the embryo. This much simpler procedure necessitates only two small holes in the shell, large enough to permit the entry of a 27-gauge needle. One hole is drilled over the air space and a second in the vicinity of the embryo. The shell membrane over the airspace is punctured to allow pressure equalisation when 0.1 ml. of inoculum is injected into the allantoic cavity. Both openings are closed with a little molten paraffin wax.

Forty-eight hours incubation usually yields allantoic fluids adequate both in volume and titre for subsequent typing.

viii. *Preservation of the Virus:* Lyophilisation provides the best method for "permanent" preservation and transport of strains. For this purpose fresh virus in infected allantoic fluid is readily freeze-dried and has good viability.

"Snap" freezing and storage in dry-ice containers is the usual method of maintaining viable stock for current use.

Egg-adapted virus will maintain its viability in infected allantoic fluid at 0 - 4° for a few weeks. Virus to be used for agglutinating purposes only may be stored at 0 - 4°, preserved with 50% Glycerol or with 0.01% Merthiolate.

Influenza is an acute self-limiting disease. As a diagnostic aid the isolation of the causal agent may be of little value except perhaps to distinguish influenza from other infectious diseases which closely simulate it. Isolations made in epidemics such as the recent "Asian Influena" outbreak can, however, be of major importance in epidemiological studies.

*A volume of about 20 mls. of sterile saline used for gargling is ejected into an equal volume of nutrient broth (Difco Nutrient Broth is excellent).

REFERENCES

1. SMITH, W., ANDREWS, C. H. and LAIDLAW, P. P. (1933). *Lancet.* **2,** 66.
2. ANDREWES, C. H., LAIDLAW, P.O. and SMITH ,W. (1934). *Lancet.* **2,** 859.
3. SMITH, W. (1935). *Brit. J. Exo. Path.* **16,** 508.
4. BURNETT, F. M. (1940). *Aus. J. Exp. Biol.* **18,** 353.
5. NIGG, C., CROWLEY, J. H. and WILSON, D. E. (1940). *Science.* **91,** 603.
6. HIRST, G. K. (1941). *Science.* **94,** 23.
7. CLARK, E. and NAGLER, F. P. O. (1943). *Aus. J. Exp. Biol. & Med. Sci.* **21,** 103.
8. HIRST, G. K. (1942). *J. Exp. Med.* **75,** 49.
9. BURNET, F. M. and BULL, D. R. (1943). *Aus. J. Exp. Biol.* **21,** 55.

SITUATION VACANT

STATUTES OF INTERNATIONAL CONGRESS OF MEDICAL LABORATORY TECHNOLOGISTS

or

(Medical Laboratory Technologists International Association)

Art. 1

NAME, LAW GOVERNING, RESIDENCE, LANGUAGES

The name of the association shall be "International Congress of Medical Laboratory Technologists" (or "Medical Laboratory Technologist's International Association").

The Association shall be governed by these Statutes in accordance with Article 60 and the following articles of the Swiss Civil Code and shall be incorporated in accordance with Article 60 of the aforesaid code.

The residence of the Association shall be Zurich, Switzerland. By decision of the General Assembly of Delegates the residence may be at any other place in Switerland or in another country.

The official languages of the Association are French and English, but those entitled to be present at General Assemblies may use their own languages and have them translated into one of the official languages at their own expense.

Art. 2

OBJECTS

The aims of the Association shall be:

(a) to afford opportunities for medical laboratory technologists to meet at stated times to confer upon questions relating to health and well-being of humanity and upon problems relating to their work;

(b) to provide a means of communication between medical laboratory technologists in different countries, to promote the general interest of medical laboratory technologists and to further friendship and understanding between the medical laboratory technologists of the world;

(c) to raise the standard of education and formation of medical laboratory technologists and offer young medical laboratory technologists opportunities of further training.

The Association excludes from its programme all political and religious discussion.

Art. 3

MEMBERSHIP

Membership shall be open to:

(a) *National Organisations,* which must consist only of medical laboratory technologists qualified according to the accepted standard of the profession in their own country. All medical laboratory technologists must be eligible for membership to these national organisations, irrespective of race, religion or opinion. Officers of these national organisations must be elected by the free vote of their members.
They may be:

1. National Associations of medical laboratory technologists which apply for membership and are accepted by the Council. Normally only one association in each country shall be approved; however, if there are in one country more than one national association of good standing, they can all be approved and will have joint membership.
2. Groups of medical laboratory technologists in countries having no association and which, having formed themselves into sections (or branches) apply for membership and are accepted by the Council, provided that only one group in each country shall be so approved.

(b) *Individual Members:* Any medical laboratory technologist who has applied direct to the Association for membership and has been accepted by the Council after due inquiries have been made. Individual members shall not have the right to vote or to hold office.

(c) *Honorary Members:* Any person who has rendered to the profession such signal service as to merit this recognition may, upon recommendation and approval by the Council be made an honorary member of the Association. Honorary members shall have no membership duties or rights.

National organisations or individuals wishing to become members shall forward an application for membership to the Executive Secretary. National Associations shall join their statutes, groups their rules, individuals their curriculum vitae and name two references.

The applications will be submitted to the Council and, if approved, the new members will be accepted subject to ratification by the next General Assembly of Delegates.

Privileges of membership will become effective and dues will be payable upon notification of provisional acceptance by the Council—dues pro rata temporis.

Art. 4

CONTRIBUTIONS

Each National Organisation and each Individual Member shall pay an annual subscription.

The National Organisations will be classified by the Council with "Units" according to their importance: up to 100 members = 1 unit; 100 to 500 members = $2\frac{1}{2}$ units; 500 to 1,000 members = 5 units; 1,000 to 2,000 members = $7\frac{1}{2}$ units; 2,000 to 5,000 members = 10 units; over 5,000 members = 15 units.

Every year the Executive Secretary has to be informed of the number of members of each National Organisation. For a transitory period and due to important reasons the Council can upon request classify a National Organisation with less units than would correspond to its members.

The amount of the annual subscription of Individual Members and of the annual unit of National Organisations will be fixed by the General Assembly of Delegates. The same amounts shall be due on January 1st of each year till they are changed by another General Assembly of Delegates.

Art. 5

POWERS OF THE ASSOCIATION

The powers of the Association shall be vested in the General Assembly of Delegates, the Council, the Executive Secretary, the Committees and the Auditors, subject to the following provisions

Art. 6

GENERAL ASSEMBLY OF DELEGATES

Those entitled to be present at General Assembles are:

(a) Delegates: Each country is entitled to send delegates to General Assemblies in the proportion of one to 200 members of its National Organisations, but at least two delegates by country. The delegates must be elected by the Assembly of the members of the National Organisations.

(b) The members of the Council.

(c) The Individual Members.

The members of the Council and the Individual Members are entitled to attend the General Assemblies and to take part in all discussions, but they have no right to vote.

The Delegates have the right to take part in all discussions and to vote. A decision is taken by a majority of those voting except where the law or these statutes state it expressly otherwise. Whenever by a

majority vote the Delegates request it, a resolution must be subsequently submitted to a vote in the National Organisations and must be admitted by a majority of these Organisations.

The Council shall call a General Assembly of Delegates with at least three months notice to the members. The notice must name the subjects of the General Assembly. With notice to the Executive Secretary at least one month before the General Assembly a National Organisation or 15 Individual Members can request a further subject to be enlisted. This is at once to be transmitted to the other members. Only the subjects enlisted may be discussed and voted on at a General Assembly.

A quorum of 20 Delegates of five different countries will be necessary for a meeting of the General Assembly.

At each General Assembly the Delegates have to present their powers to represent their National Organisation and the Individual Members have to identify themselves.

An *Ordinary General Assembly* of Delegates takes place if possible every year, but at least every two years. The date and place are fixed by the previous General Assembly, this upon invitation of a National Organisation. Chairman of an Ordinary General Assembly is the

president of the inviting National Organisation, if there are more than one he will be chosen by the previous General Assembly.

The Council can call at any time an *Extraordinary General Assembly* of Delegates if so needed. He must do so if it is requested by at least five National Organisations or 50 Individual Members. Chairman of the Extraordinary General Assemblies is the president of the Council.

A General Assembly of Delegates can decide on any subjects which are not expressly in the competence of others. It shall especially—

Ratify the membership of new members;

Accept the annual report of the Council, the annual account and the report of the auditors;

Discharge the Council, the Executive Secretary, the Committees and the Auditors of their responsibility for the past years;

Elect the Council, the Executive Secretary and the Auditors;
Amend the Statutes, dissolve and liquidate the Association.

Art. 7

THE COUNCIL

The Council consists of a President, a Vice-President for public relations to non-European countries, an Executive Secretary, a Recording Secretary and a Treasurer. Its members must be members of National Organisations and represent at least four separate countries.

The members of the Council shall be elected by the General Assembly for a period of two years and shall be eligible for re-election for further terms. The Council shall in any case hold office till another General Assembly is able to make another election. In the case of resignation or death of one member during a term the others shall appoint a successor for the rest of the term.

The Council meets at least once each year upon invitation of its President. Notice of the meeting must be given at least 14 days in advance and enlist the subjects to be discussed. Only subjects enlisted can be discussed. The Council can take its decisions also in writing if opportunity is given to every member to discuss the matter and to vote. Every decision is taken by the majority of those voting.

The Council carries out the decisions of the General Assemblies of Delegates. It deals with current business and represents the Association. It decides upon the signature of the Association. It issues by-laws to the present statutes.

Art. 8

THE EXECUTIVE SECRETARY

He deals with current business in the interval between the meetings of the Council. He shall especially—

receive all correspondence and all payments made to the Association, study them and forward them with his commentary to the other members of the Council concerned;

keep all records and documents of the Association;

inform the National Organisations of any matters affecting the Medical Laboratory Technologists and the Associations;

prepare the meetings of the Council and its written decisions—

prepare the meetings of the General Assembly of Delegates;

prepare the annual report dealing with the activities of the Association and also with any other matters of interest connected with the Medical Laboratory Technologists.

Art. 9

COMMITTEES

Any standing or special (ad hoc) committee may be set up by the Council as required. The Executive Secretary shall be ex officio member of all committees.

Art. 10

AUDITORS

The General Assembly shall elect two auditors who hold offices for two years or appoint a trustworthy Swiss Auditing Company.

They shall audit the accounts of the Association before each General Assembly, but at least every calendar year and present their report to the General Assembly in writing.

Art. 11

AMENDMENTS TO THE STATUTES, DISSOLUTION OF THE ASSOCIATION

The present Statutes can only be altered at a General Assembly of Delegates. The Association can only be dissolved and liquidated at a specially called General Assembly of Delegates.

Motions concerning amendments of the Statutes or dissolution of the Association shall be sent to the Executive Secretary at least within one month to the National Organisations, i.e., at least five months before the date of the General Assembly.

At such General Assembly at least half of the National Organisations must be represented. A two-third majority of those present and voting will be necessary.

A change of the aims of the Association or its dissolution must moreover subsequently be submitted to a vote in the National Organisations and must be approved by a majority of these Organisations.

The present Statutes were accepted by the Initial General Assembly held in Bristol in September, 1958.

For the Assembly.

THE CHAIRMAN.

INTERNATIONAL CONGRESS OF MEDICAL LABORATORY TECHNOLOGISTS

For the interest of members we publish the draft constitution of the above organisation. Unfortunately this Society is unable to send delegates to the Congress of the Institute of Medical Laboratory Technology which is being held in Bristol, England, as we go to press, but we hope that members will not hesitate to send criticisms and/or suggested amendments to this constitution so that they may be forwarded to Miss Pletscher in Zurich.

An international body representing medical technologists is very necessary and it is hoped that all medical technologists will support its aims by joining their own societies and organisations and playing their part in the furthering of interest in the problems of medical technology which is arising the world over.

G. W. WIKELEY,
Hon. General Secretary.

PRIZE IN PARASITOLOGY

The Director of the Amoebiasis Research Unit, C.S.I.R./N.P.A./Univ. of Natal announces :—

(1) Entries are invited from members of the Society for a prize to be awarded for an essay not exceeding 2,000 words on the subject :—

"The Common Round Worm."

(2) Both qualified and student technologists are eligible.

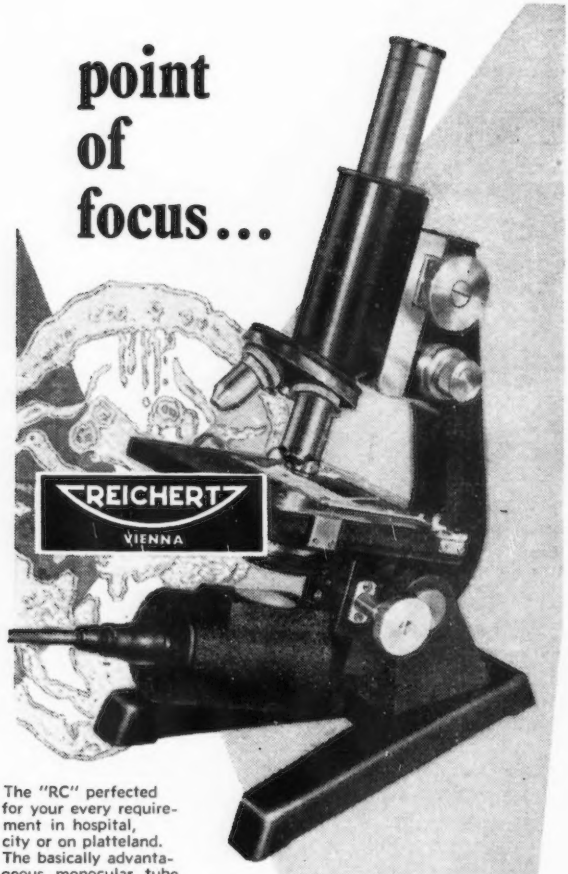
(3) All entries shall be the property of the S.A. Journal of Medical Laboratory Technology.

(4) The entries shall be judged by a panel of three including the donor.

(5) The adequacy of documentation and illustration shall be taken into account. Such illustrations need not be drawn by the author, but acknowledgment must be made.

(6) Entries shall be in the hands of the General Secretary by 31st December, 1958.

point of focus...



The "RC" perfected for your every requirement in hospital, city or on platteland. The basically advantageous monocular tube means optimum illumination even with daylight at high magnification.
Reasonably priced standard combinations for the daily round — or adapted for multipurpose use . . . **Darkground; Photomicography; Phase-contrast; Biological Warmplate** (keeps specimens at blood temperature).
All **Reichert** microscopes sold under full guarantee !

PROTEA HOLDINGS LTD.
NUCLEONICS DIVISION

42, Faraday Street,
JOHANNESBURG.
Telephone 33-2211. P.O. Box 7793.

Westoby 1847